THE EFFECT OF LEAF AGE AND ABSCISSION ZONE FORMATION ON THE ABSORPTION AND TRANSLOCATION OF 2,4,5-TRICHLOROPHENOXYACETIC ACID BY BLACKJACK OAK (QUERCUS MARILANDICA MUENCHH.)

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CHAPTER I

INTRODUCTION

Millions of acres of land are occupied by brushy plant species in the United States. Oklahoma has an estimated 10 million acres of this undesirable vegetation according to Elwell et al. (13) /1.

A dense brush canopy of woody vegetation greatly reduces the growth potential of desirable grasses, legumes, and trees in non-cultivated areas. Some of this land is rough, rocky, and unsuited for the growth of grass. Much of it, however, is well suited to profitable production of timber and nut-bearing trees and desirable forage grass species. Efforts are being made to reclaim this land through new and more efficient range management practices.

The phenoxy-type herbicides, introduced in the early 1940's, have proved to be fairly effective as industrial herbicides. At present, there seems to be no other material that will compare favorably with the phenoxy compounds in their effectiveness and ease in handling. However, 2,4-dichlorophenoxyacetic acid, hereafter referred to as 2,4-D, and 2,4,5-trichlorophenoxyacetic acid, hereafter referred to as 2,4,5-T, are not consistent in their effects on the brushy species of this area. The level of control of blackjack oak (*Quercus mari-*

/1 Figures in parenthesis refer to Literature Cited.

landica Muenchh.) and post oak (Quercus stellata Wang.) has tended to vary from year to year and also within seasons. The present work was initiated to determine some of the specific causes of this variability.

The observations of Elwell (14) showed that 2,4,5-T is often more effective for the control of blackjack oak when applied in the early spring. The recommended treatment dates of May 15 to July 15 suggest that environmental conditions and possibly the age or physiological condition of the leaf are important factors in determining the effects of 2,4,5-T under field conditions. Since the absorption and translocation of the herbicide are two of the most critical factors in determining the toxicity of 2,4,5-T, these factors were studied in blackjack oak in relation to leaf age or date of application.

Other observations which have been made by Elwell (15) and which may have some relationship to the degree of toxicity of 2,4,5-T toward blackjack oak concerns the rapid formation of abscission zones in the pulvenoid leaf tissues of treated plants. After treatment, many of the leaves of treated plants have been observed to absciss and fall within a few days. It seemed possible that the abscission zone in the leaf might be formed rapidly enough to prevent adequate translocation of 2,4,5-T from the leaf to stem and root tissues and thereby lower the toxicity of the herbicide. Studies were initiated to determine the rate of abscission zone formation in treated leaves at various dates throughout the season.

CHAPTER II

LITERATURE REVIEW

To exert herbicidal action on a plant, a chemical applied to the foliage must first be absorbed by the leaf or stem. Recent evidence reported by Dybing and Currier (12) has shown that absorption by leaf tissue may occur both through the cuticle and through open stomates and that the route taken for absorption will depend upon the type of chemical and carrier being used.

The stomatal entry of herbicides in oil solutions is well known reports Currier and Dybing (11) and apparently is not a controversial issue. However, the stomatal entry of aqueous solutions is an issue in which conflicting evidence has been apparent. While Cook and Boynton (6), Gustifson (16) and Skoss (34) have reported that aqueous solutions readily enter open stomates, Crafts (10), Scott et al. (33), Thimann (36) and Weaver and DeRose (39) have reported that there was little entry through the stomates.

The position taken by van Overbeck (37) and Leonard and Crafts (21) suggesting that aqueous solutions enter stomates only when sufficient surfactant is present is probably the more realistic one. The recent work of Dybing and Currier (12) clearly shows that a surfactant was essential for the stomatal entry of a number of compounds and that this type of absorption probably accounts for most of the absorption of 2,4-D and 2,4,5-T in most species of plants. However, penetration of the cuticle does occur as was shown for bean leaves by Dybing and Currier (12) and a difference in type of cuticle could well bring about variation in the effectiveness of herbicides for some types of plants.

Electron microscope studies by Mueller et al. (25) have illustrated that variations occur in the wax development on the leaf surfaces of different species during the changing stages of development and that environmental factors may influence the type of development as reported by Juniper (20). Wax patterns in growing leaves suggest that the upper wall of epidermal cells enlarge at the cell margins. Possibly the greater susceptibility of growing leaves to herbicides is related to a permeable, immature zone in the cuticle through which the herbicide can penetrate rapidly according to Schieferstein and Loomis (32).

A number of environmental factors may influence absorption. Hauser (17) reported that less 2,4-D was absorped by plants grown under moisture stress. However, Pallas (26) states that soil moisture did not influence the absorption of 2,4-D by bean plants when the plants were subjected to short periods of drought. The work by Pallas (27) has clearly shown that relatively high temperatures as well as high humidities are favorable for the translocation of 2,4-D from the treated leaf to the epicotyl tissue in bean plants. It was suggested that these results may be correlated with absorption of 2,4-D as regulated by the degree of stomatal opening.

Some effects of temperature on the absorption of 2,4-D have been reported by Barrier and Loomis (2) and Hauser (17) who have shown that high temperatures favor absorption or possibly translocation. Rice (28) showed that absorption of 2,4-D was increased by increasing the temperature.

Behrens (4) stated that the phytotoxicity of the triethanolamine salt of 2,4,5-T decreased as the size of the droplets and the volume of the spray were reduced. He attributed this to restricted absorption. Blackman and Robertson-Cuninghame (5) found a lower absorption of 2,4-dichloro-5-iodophenyoxyacetic acid when the plants were kept at a low light intensity after application. After working with bean seedlings Hay and Thimann (19) stated that 2,4-D may be absorbed by, but not translocated from leaves in darkness.

After absorption the herbicide must next be translocated to the stems and roots for maximum toxic effects. The initial toxic effects of the herbicide may determine whether transport can readily occur. After experimenting with Dichrostachys, Hay (18) stated that large amounts of 2,4-D and 2,4,5-T were prevented from moving down the plant through living tissue because the herbicides disrupted the requisite transport mechanism when they were applied in excessive amounts. The effectiveness of herbicide applications may also depend to a large extent upon the movement of assimilates in the phloem according to Crafts (7), (8) and Leonard and Crafts (21). Metabolic energy is supplied to the phloem in the form of carbohydrates, products of the photosynthetic activity of the leaves. Zimmerman (41) reported these carbohydrates may provide, at the same time, the energy requirements for the movement of organic substances such as 2,4,5-T in the phloem cells. It has been shown repeatedly by Barrier and Loomis (2), Mitchell and Brown (24), Rohrbaugh and Rice (30) and Weintraub and

Brown (40) that the translocation of large amounts of 2,4-D from leaf tissue requires a high carbohydrate level in the plant tissue.

The rate of phloem translocation is often extraordinarily high. According to Mason and Maskell (22) it can be as much as 40,000 times the rate of sugar diffusion in water. Some reports gave the impression that 2,4-D moves relatively freely in plants, accompanying foods in their passage from regions of their synthesis to regions of their utilization (7, 8, 21). In a comparative study of several herbicides, Crafts and Yamaguchi (9) reported that 2,4-D and 2,4,5-T are somewhat restricted in their distribution and this was attributed to the structure of the molecule.

There are also a number of environmental factors which influence translocation of the phenoxy herbicides in plant tissue. Pallas (26) has shown that the soil moisture level causes marked effects on translocation of 2,4-D and Basler et al. (3) have shown that soil moisture may affect translocation by altering the relative turgidity of plant cells. When turgidity of leaf cells decreased to a critical minimum only trace amounts of 2,4-D were translocated. Pallas (27) has shown that high temperatures as well as high humidities are favorable for translocation. However, it was stated that these effects may have been due to effects on absorption. The data of Barrier and Loomis (2) and Hauser (17) also suggests that high temperatures favor translocation. Rice (28) concluded that, although the absorption of 2,4-D is increased by increasing temperatures, translocation is relatively unaffected by temperature. Rice and Rohrbaugh (29) have, however, shown that temperature may be a very critical factor in the translocation of 2,4-D in destarched plants. In destarched plants the translocation of 2,4-D was significantly inhibited by subjecting the plants to high temperatures during the pretreatment period.

CHAPTER III

MATERIALS AND METHODS

The test plants used in this study were blackjack oak growing in their natural habitat. The study was conducted in the S $\frac{1}{2}$, T 19 N, R 1 E located in the Lake Carl Blackwell area, west of Stillwater, Oklahoma. This tract was native grass pasture containing several tree species. The dominant tree species of the area was Quercus marilandica Muenchh. The soil was Darnell and/or Stephenville sandy loam (1).

Trees approximately 10 feet in height were selected to insure uniformity of plant size for all treatments. Single plants spaced several feet apart were selected and tagged. In some instances, this made it necessary to remove the closely competitive trees.

The absorption and translocation of 2,4,5-T was studied by tracing the movement of C^{14} contained in the carboxyl group of the herbicide, 2,4,5-T-I-C^{14}. Three trees were selected for each treatment and treatments were made at 2 to 4 week intervals, from May 2, 1960, when leaves were about two-thirds developed, until October 4, 1960. Six leaves, on each tree, were treated with the 2,4,5-T-I-C^{14}. Six treated leaves, two from each tree, were removed at 12, 24, and 36 hours after treatment and each was analyzed separately for C^{14}. In order to simulate the conditions used for spraying for the control of brush species in this area, as nearly as possible, the entire tree, except for the treated leaves, was sprayed with a nonlabeled solution of 2,4,5-T. To prevent the leaves which were selected for treatment from being wet with the nonlabeled 2,4,5-T, each of these leaves were enclosed for about 10 minutes in small plastic bags while the nonlabeled 2,4,5-T was being applied. The 2,4,5-T-I-C^{14} was applied immediately after removing the plastic bags.

The 2,4,5-T-I-C^{14} and the nonlabeled 2,4,5-T were applied as water emulsions of the butoxy ethanol ester of 2,4,5-T. The nonlabeled emulsions were prepared by mixing 1 part 2,4,5-T (4 pounds acid equivalent per gallon) and 99 parts water (14). A 3-gallon knapsack hand sprayer was used to apply the emulsion. Foliage of the trees was thoroughly wet. All applications were made in the early morning.

The 2,4,5-T-I-C^{14} emulsion was prepared at the beginning of the growing season and was stored at about 4° C. Each leaf treatment contained approximately 36 μg. of 2,4,5-T acid equivalent with a specific activity of .95 mc/mM. The microcuries of C^{14} in each of two aliquots of the emulsion equal to one leaf treatment, about .13 μc. in .01 ml. of emulsion, was determined at the time of each treatment date and was found to vary slightly throughout the season as shown in Appendix Table 1. This variability may have been caused by inconsistancies in the emulsion or to evaporation of the solution and breakdown of the 2,4,5-T during the season of application. After spraying with nonlabeled 2,4,5-T and removal of the bags a micropipette was used to place a 10 microliter drop of 2,4,5-T-I-C^{14} on the upper surface of each of the six leaves. The labeled herbicide was placed to the left of the midrib and on the apical side of the left lateral vein in an area of minor veination.

Following the collection of samples at 12, 24, and 36 hours after treatment the unabsorbed 2,4,5-T-I-C^{14} was determined by washing the treated area of each leaf with 25 ml. of 80% ethyl alcohol and determining the C^{14} in each sample. Absorption in this study is represented by the value obtained by subtracting the leaf wash C^{14} from the total C^{14} applied at the time of treatment. The C^{14} in each washed leaf was also determined and translocation in this experiment is represented by the values obtained by subtracting the leaf C^{14} and the wash C^{14} from the total C^{14} applied.

Aliquots of the washed leaf were analyzed after homogenizing the leaf in a Virtus blender in 15 ml. of 80% ethanol. The homogenizer shaft and container were washed with 10 ml. of alcohol to make 25 ml. of leaf extract. The washes and extracts were put in test tubes and sealed with rubber stoppers. The test tubes were stored at minus 15^{o} C. until completion of the analysis. Two ml. of leaf wash and 2 ml. of leaf extract was analyzed for each of the leaves that were collected. One drop of saturated NaOH was placed in each oxidization chamber before pipetting the aliquots of leaf wash or leaf extract. The samples were then dried by use of a stream of forced hot air. The C^{14} of each sample was converted to $C^{14}O_2$ by wet combustion of the residues preformed according to the method of Van Slyke et al. (38). The isotope in each aliquot was collected in a 250 ml. gas ionization chamber and measured with a Nuclear-Chicago Dynacon Electrometer.

Both absorption and translocation values may be somewhat high since the technique employed does not account for evaporation and destruction of 2,4,5-T on the leaf surface. The possibility also exists that some of the 2,4,5-T was converted to other substances in the leaf tissue and was translocated as such.

Records of the soil moisture and the daily rainfall were kept for the area of this experiment throughout the season under consideration. The data for daily rainfall was obtained from the Outdoor Hydraulic Laboratory, Stillwater, Oklahoma, which is located in the near vicinity of the test plots. Soil moisture determinations were made at the time of each treatment. Soil samples were taken with a 1 inch by 4 foot sampling tube. Three samples were taken at 1, 2, 3 and 4 foot depths. One sample was taken near each of the treated trees. The samples were dried at 105^{o} to 110^{o} C. according to Millar and Turk (23) with the moisture content being determined on a dry weight basis.

Pulvenoid tissues were collected from treated and untreated leaves for the study of abscission zone formation as affected by 2, 4,5-T. The 2,4,5-T-I-C^{14} treated leaves were removed from the trees with a sharp razor blade in such a manner that all of the pulvenoid tissue remained with the leaf. This pulvenoid tissue was then removed from the leaf and preserved for analysis. Samples for these analyses were taken during the 1959 and the 1960 growing season. During 1959, two pulvenoid tissues and the corresponding leaves were taken at random from the tagged leaves of each of three trees at 24, 72, and 120 hour intervals after the 2,4,5-T-I-C^{14} applications. The 1960 collections were made at 12, 24, and 36 hours after treatment. The pulvenoid tissue was killed and fixed in formalin, chromic and acetic acids, and water (Craf's solution) according to Sass (31). Slides were prepared by imbedding, sectioning, staining with safranin and fast green, and mounting according to the schedule outlined by Sass

(31). The slides were analyzed microscopically with the oil emersion lens and representative microphotographs were prepared.

The split-plot design was used to facilitate statistical analyses of the 2,4,5-T-I-C^{14} not absorbed, the accumulation in the leaf, the absorption and the translocation of one rate of herbicide applied to blackjack oak leaves. The main plot effect was declared to be treatment dates and the time of treatment or collection periods were in the sub plots. Due to unequal spacing, the sums of squares for the linear, quadratic, cubic, quartic and residual trends of the treatment dates were obtained by using the abbreviated Doolittle procedure. Statistical analyses were performed as described by Snedecor (35).

CHAPTER IV

RESULTS

Through observation of the treated leaves it was found that a burning effect or contact injury was readily noticeable at the point of herbicide application early in the season. This browning effect occurred on all treated leaves during the 12 hours following applications on May 2 and May 17. Similar, but apparently less severe effects were noted on 50% of the leaves treated June 3. The browning of tissue was not observed on any of the leaves treated at a later date.

The data for the absorption of 2,4,5-T-I-C^{14} in blackjack oak are shown in Figure 1. The absorption of 2,4,5-T-I-C^{14} varies considerably during the season. Absorption at 12 hours after treatment was fairly high at the beginning of the study in early May. There was a sharp decrease in absorption until about the middle of June at which time a gradual increase in rate of absorption began and continued until mid-August. At the middle of August absorption begins a second decrease, which continues until the end of the growing season. The curves for the 24 and 36 hour treatment effects show similar trends, however, absorption increased at 24 and 36 hours after treatments over the 12 hour treatment for most of the treatment dates. This statistical analysis in Appendix Table II showed a significant

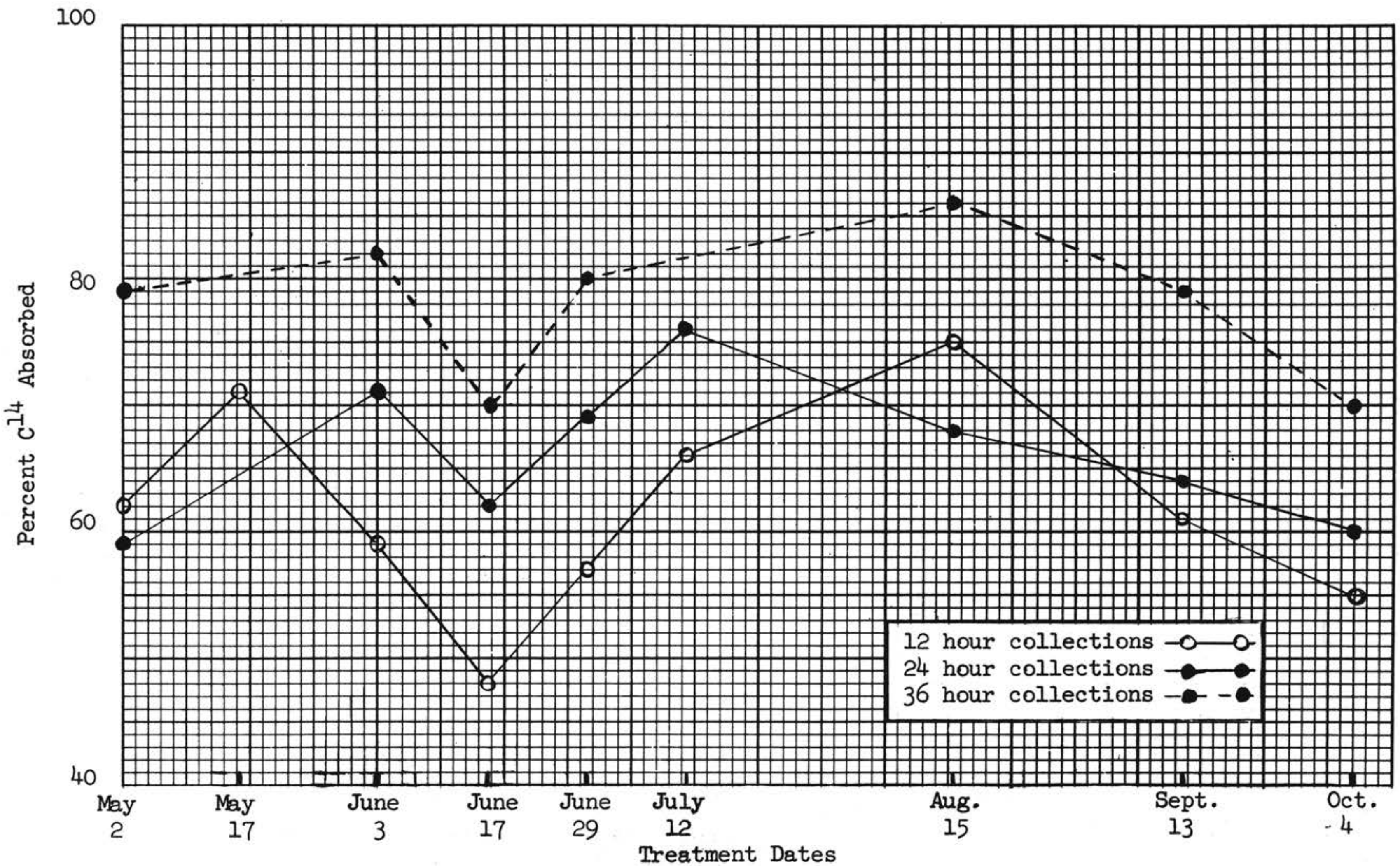

Figure 1. Percent of the total applied C^{14}-labeled 2,4,5-T absorbed at 12, 24 and 36 hours after foliar applications to blackjack oak. The individual values represent averages for six determinations.

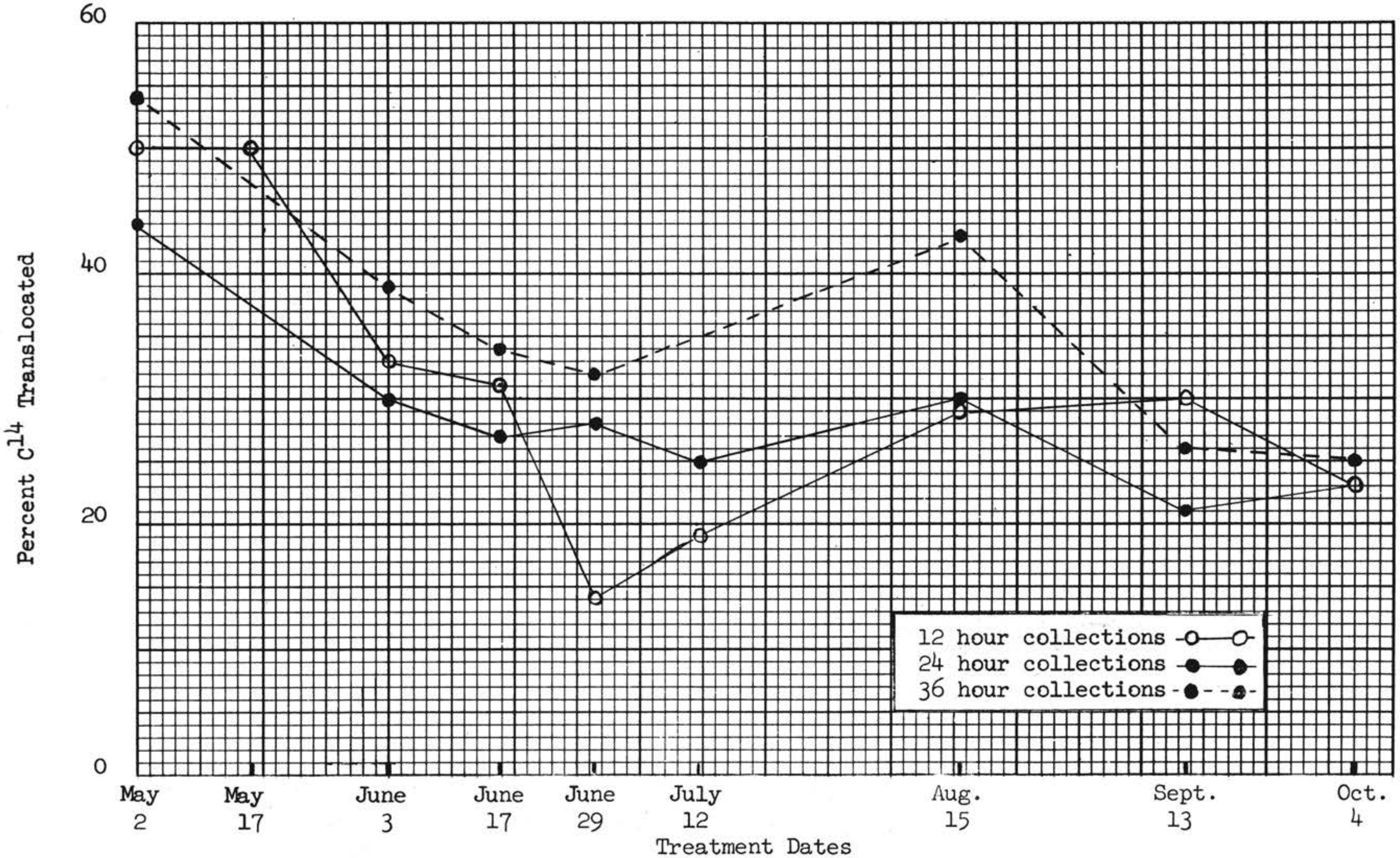

Figure 2. Percent of the total applied C^{14}-labeled 2,4,5-T translocated from the leaf at 12, 24 and 36 hours after foliar applications to blackjack oak. The individual values represent averages for six determinations.

linear and quadratic response for absorption at 12, 24, and 36 hour collections. In addition, the various treatment dates were shown to exhibit a cubic response that was significant at the 5% confidence level.

The data for the translocation of 2,4,5-T-I-C^{14} are shown in Figure 2. Translocation for the 12 hour treatments was fairly high in the early spring but decreased rapidly until about the first of July. After this, the translocation rates increased slowly until near the end of the growing season. The curves for the 24 and 36 hour treatments show similar trends. The analysis of variance showed a significant linear, quadratic and cubic response for the various treatment dates during the season (Appendix Table III) and Orthogonal comparisons indicate significance at the 1% level for the 12, 24, and 36 hour collections. The data for May 17 and July 12 was considered of no statistical value. This loss of data was due to the occurrence of rain between the 12 and 24 hour and the 24 and 36 hour collections for the respective dates.

As shown in Figures 1 and 2, the absorption and translocation for the 12 hour treatments are fairly similar with increases and decreases occurring at about the same dates throughout the season. However, translocation appears to decrease more drastically during the summer months of July and August. This resulted in the accumulation of more 2,4,5-T-I-C^{14} in the leaves during this part of the season, as is shown in Figure 3. The statistical analyses for this data are shown in Appendix Table IV.

The data for soil moisture are given in Appendix Table V. Soil moisture decreased at the 1, 2, 3 and 4 foot levels during the summer

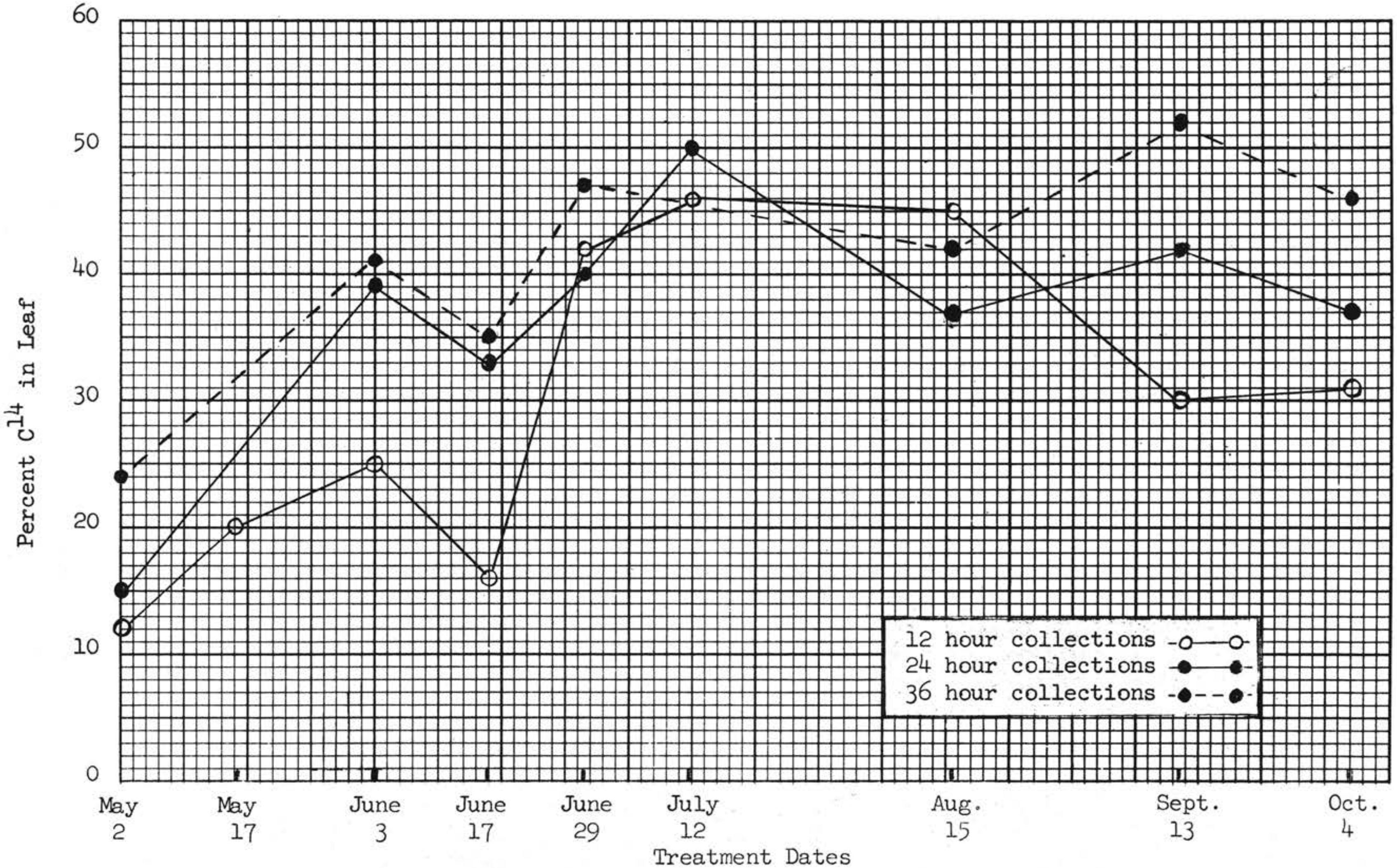

Figure 3. Percent of the total applied C^{14}-labeled 2,4,5-T accumulated in the leaf tissue at 12, 24 and 36 hours after applications to blackjack oak. The individual values represent averages for six determinations.

and fall months. The decrease was especially noticeable at the 1 foot level. No decreases in soil moisture were apparent at any level during the months of May and June. However, this is the time during which the large reduction in translocation and absorption occurred. The data for daily rainfall are given in Table VI. The data showed that considerable amounts of rainfall occurred during the months of May, June and July. The months of May and July were quite high when compared with the long time mean.

Figures 4 and 5 show photomicrographs depicting the typical results obtained from the study on abscission zone formation. Although extensive slides were prepared throughout the 1959 season, of the various intervals after treatment, no indications were apparent that either cell wall hydrolysis, cell division or clogging of phloem cells with tallous material was occurring.

It was evident from the ease of removal of treated leaves from the tree at the abscission zone and from the rate of leaf fall from the trees during the later hours of treatment that leaf abscission processes were occurring. However, these effects were apparently limited to the middle lamella and it is not known to what extent they might have influenced the rate of translocation of 2,4,5-T-I-C^{14} from the leaf.

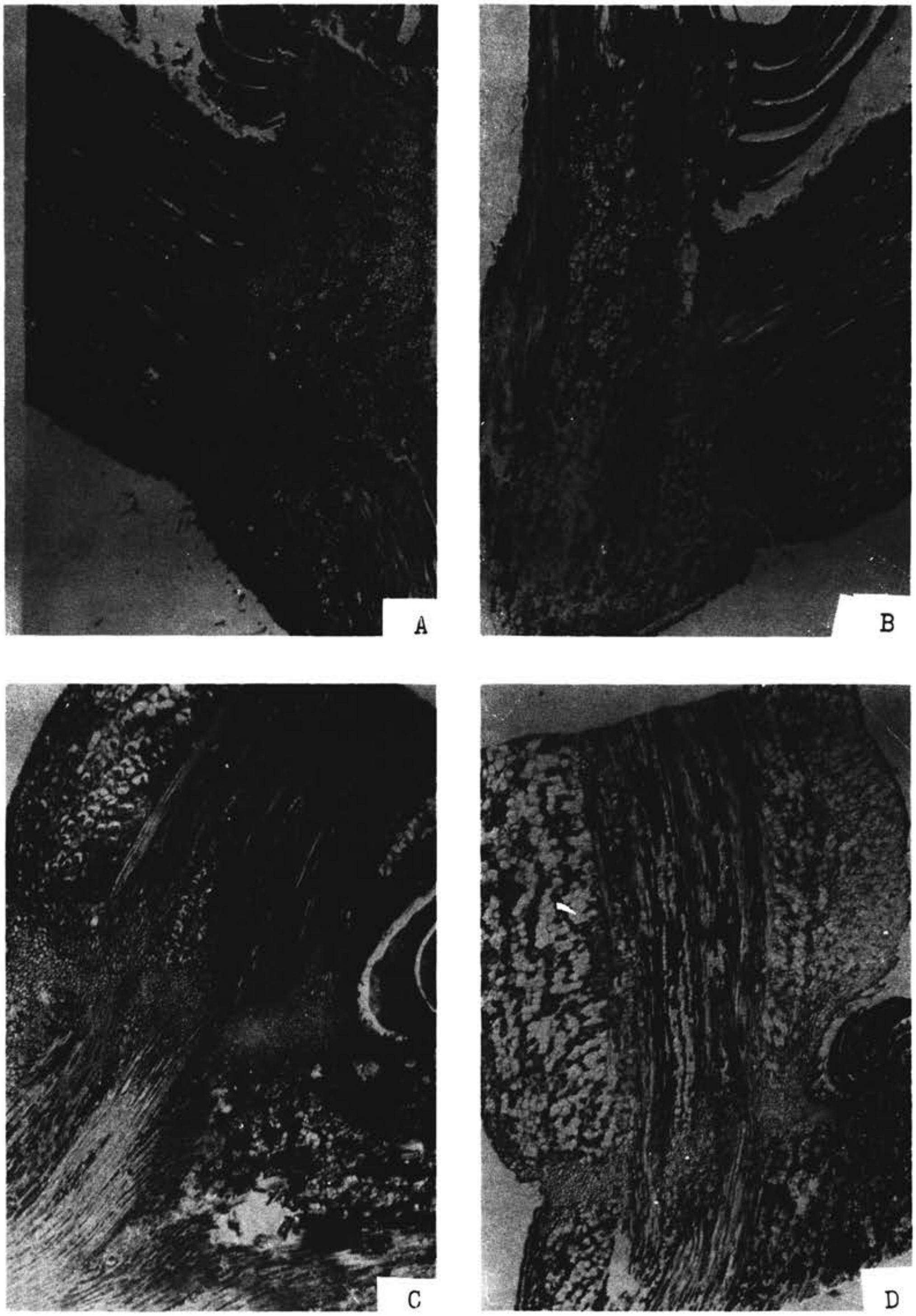

Figure 4. Longisection photomicrographs of pulvenoid tissue preceding and following 2,4,5-T applications on blackjack oak. A, preceding treatment; B, 24 hours after treatment; C, 72 hours after treatment; D, 120 hours after treatment; 25X.

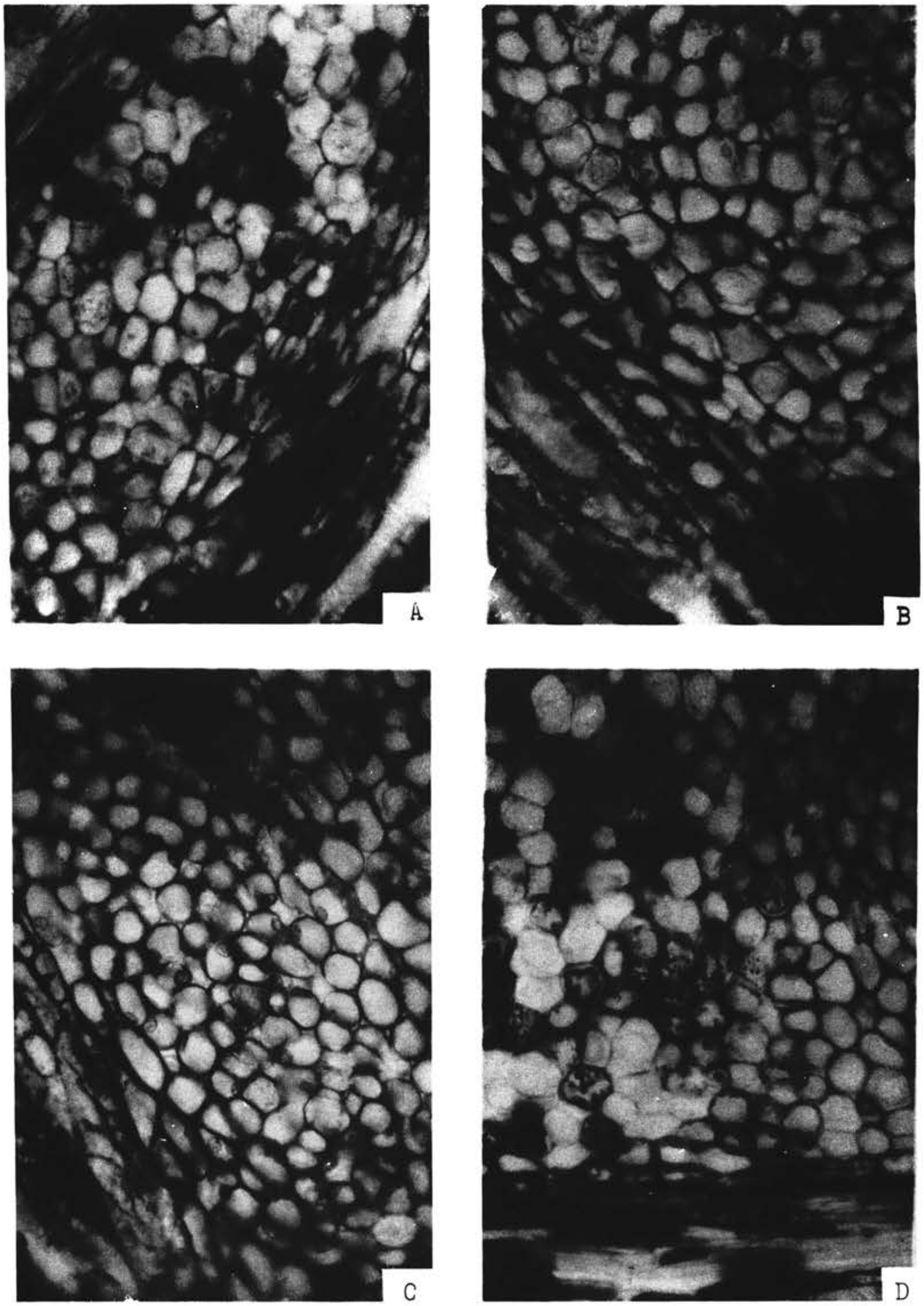

Figure 5. Longisection photomicrographs of abscission layer preceding and following 2,4,5-T applications on blackjack oak. A, preceding treatment; B, 24 hours after treatment; C, 72 hours after treatment; D, 120 hours after treatment; 350X.

CHAPTER V

DISCUSSION

The absorption and translocation of 2,4,5-T-I-C^{14} in blackjack oak were both fairly high in the early spring. There was a rapid decrease in absorption and translocation occurring in May and June so that during the summer months of July and August the absorption and translocation occurred at comparatively low rates. It has been noted that July and August are usually very unfavorable times of the year for killing blackjack oak in this area (14). Apparently, both absorption and translocation may be factors responsible for the low degree of kill during these months. However, translocation appears to be the more important factor since it seems to be reduced during July and August to a greater extent than absorption. Both absorption and translocation increase during the early fall months of August and September and it has been observed that brush may be successfully controlled, at times, by applications during the late growing season.

The sharp reduction of absorption and translocation occurred during May and June while the soil moisture level was still high, which indicates that water stress was not an important factor in these reductions. However, it has been shown that water stress may exist in plants while the soil moisture level is still fairly high and that this water stress in leaf tissue will drastically inhibit the translocation of 2,4-D in bean plants (3). It appears possible that water stress could exist in trees during days when the weather is such that the temperature is high and the humidity is low. Humidity has also been shown to affect the rate of absorption (27) with a high humidity apparently being more favorable. It does not appear logical to assume that the decrease in absorption and translocation which occurs with the approach of summer is attributed to temperature effects. It has been shown that increasing temperatures tend to favor absorption of 2,4-D in some plants (2, 16, 27, 28).

The age of the treated leaf may be a critical factor in absorption and translocation in these experiments. Changes which affect absorption may occur with the cuticle development. It appears highly probable that metabolic changes occur within the leaf, upon aging, which could influence translocation in a number of ways.

CHAPTER VI

SUMMARY AND CONCLUSIONS

The absorption and translocation of C^{14}-labeled 2,4,5-T was determined on blackjack oak at 12, 24, and 36 hours after foliar treatments on various dates throughout the growing season.

Absorption was followed by determining the non-absorbed 2,4,5-T, which was removed from the leaf by washing with 80% ethanol, and subtracting the value from the total 2,4,5-T applied to the leaf.

Translocation values were determined by analyzing both the leaf and the leaf wash and subtracting these values from the total 2,4,5-T applied to the leaf.

Also, an analysis of the rate of leaf abscission zone formation was made in an attempt to determine if this factor might influence the amount of 2,4,5-T translocated from the leaves of these plants.

The absorption and translocation of 2,4,5-T varied considerably in these plants throughout the season. Absorption and translocation were both high during the early spring but there was a sharp decrease during May and June so that both were low during the midsummer month of July. Translocation appears to undergo a slightly greater decrease than absorption. There was a gradual increase in absorption and translocation which lasted through August and September until near the end of the growing season. It was concluded that both absorption and translocation of 2,4,5-T may be factors which are responsible for the low degree of kill of blackjack oak by the herbicide when applied during the summer months of July and August.

There were obvious processes of abscission zone formation occurring in treated plants since leaf fall was fairly high after treatment. Only slight pressures were necessary to remove treated leaves from the stem by separation in the pulvenoid section of the leaf. However, these effects were not apparent upon microscopic examination of the pulvenoid tissues. There were no indications of whole cell wall hydrolysis, cell division or clogging of phloem cells with tallous formations. It was not possible to determine if abscission zone formation was influencing the translocation of 2,4,5-T from the leaf tissue.

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APPENDIX

TABLE I. THE MICROCURIES OF C^{14} APPLIED AS 2,4,5-T-I-C^{14} PER LEAF ON VARIOUS TREATMENT DATES THROUGHOUT THE SEASON. THE INDIVIDUAL VALUES REPRESENT AVERAGES FOR TWO DETERMINATIONS.

May 2	May 17	June 3	June 17	June 29	July 12	Aug. 15	Sept. 13	Oct. 4
0.124	0.130	0.131	0.127	0.132	0.128	0.119	0.113	0.114

TABLE II

ANALYSIS OF VARIANCE OF THE ABSORPTION OF 2,4,5-T-I-C^{14} 1/ BY BLACKJACK OAK LEAVES AT 12, 24, AND 36 HOURS AFTER TREATMENT

Source of Variation	D.F.	Sum of Squares	Mean Square	F Value
Total	125	24,195.55		
Dates	6	3,209.70		
Linear (1)		(2.91)	2.91	0.01
Quadratic (1)		(496.37)	496.37	3.15
Cubic (1)		(1,174.22)	1,174.22	7.45*
Quartic (1)		(9.09)	9.09	0.05
Residual (2)		(1,527.11)	763.55	4.85*
Trees in Dates	14	2,205.00	157.50	
Collection Periods	2	7,515.96		
Linear (1)		(6,992.50)	6,992.50	118.59**
Quadratic (1)		(523.46)	523.46	8.87**
Collection Periods X Dates	12	1,233.00	102.75	1.74
Collection Periods X Trees in Dates	28	1,650.99	58.96	
Sample Duplicates	63	8,380.90	133.03	

1/ Determined by subtracting the activity in the leaf wash from the amount of C^{14} applied per leaf.

* Indicates significance at the 5% level of confidence.

** Indicates significance at the 1% level of confidence.

TABLE III

ANALYSIS OF VARIANCE OF THE TRANSLOCATION OF 2,4,5-T-I-C^{14}[1/]
FROM THE LEAF OF BLACKJACK OAK AT 12, 24 AND 36
HOURS AFTER TREATMENT

Source of Variation	D.F.	Sum of Squares	Mean Square	F Value
Total	125	26,479.60		
Dates	6	8,372.87		
Linear (1)		(4,539.21)	4,539.21	22.70*
Quadratic (1)		(1,329.22)	1,369.22	8.05**
Cubic (1)		(1,636.35)	1,636.35	9.62*
Quartic (1)		(3.64)	3.64	0.02
Residual (2)		(824.45)	412.22	2.42
Trees in Dates	14	2,386.28	170.04	
Collection Periods	2	1,284.42		
Linear (1)		(782.63)	782.63	4.75**
Quadratic (1)		(501.79)	501.79	3.05
Collection Periods X Dates	12	1,554.84	129.57	0.79
Collection Periods X Trees in Dates	28	4,603.37	164.44	
Sample Duplicates	63	8,277.82	131.39	

1/ Determined by subtracting the leaf wash plus the leaf extract from the amount of activity applied per leaf.

* Indicates significance at the 5% level of confidence.

** Indicates significance at the 1% level of confidence.

TABLE IV

ANALYSIS OF VARIANCE OF THE 2,4,5-T-I-C^{14}1/ ACCUMULATED IN THE LEAF BY BLACKJACK OAK AT 12, 24 AND 36 HOURS AFTER TREATMENT

Source of Variation	D.F.	Sum of Squares	Mean Square	F Value
Total	125	29,774.18		
Dates	6	9,641.48		
Linear (1)		(4,318.62)	4,318.62	20.13*
Quadratic (1)		(3,524.84)	3,524.84	16.43*
Cubic (1)		(36.70)	36.70	0.17
Quartic (1)		(23.62)	23.62	0.11
Residual (2)		(1,737.70)	868.84	4.04*
Trees in Dates	14	3,003.28	214.52	
Collection Periods	2	3,096.55		
Linear (1)		(3,096.42)	3,096.42	22.91**
Quadratic (1)		(0.03)	0.03	0.00
Collection Periods X Dates	12	1,993.40	166.11	1.23
Collection Periods X Trees in Dates	28	3,773.75	134.77	
Sample Duplicates	63	8,265.72	131.20	

1/ Determined from the percent of C^{14} in the homogenized leaf extract.

* Indicates significance at the 5% level of confidence.

** Indicates significance at the 1% level of confidence.

TABLE V

PERCENT SOIL MOISTURE ON A DRY WEIGHT BASIS AT VARIOUS DEPTHS IN THE TREATMENT AREA

Date	Depth In Feet *				
	1	2	3	4	Av.
May 2	14	14	15	16	14.75
May 17	12	14	15	16	14.25
June 3	14	14	15	16	14.75
June 17	13	13	15	17	14.50
June 29	12	12	14	15	13.25
July 12	9	10	10	15	11.00
Aug. 15	7	11	13	13	11.00
Sept. 13	9	10	10	13	10.50
Oct. 4	9	10	10	14	10.75

* The average moisture content as determined from three samples at each of the depths.

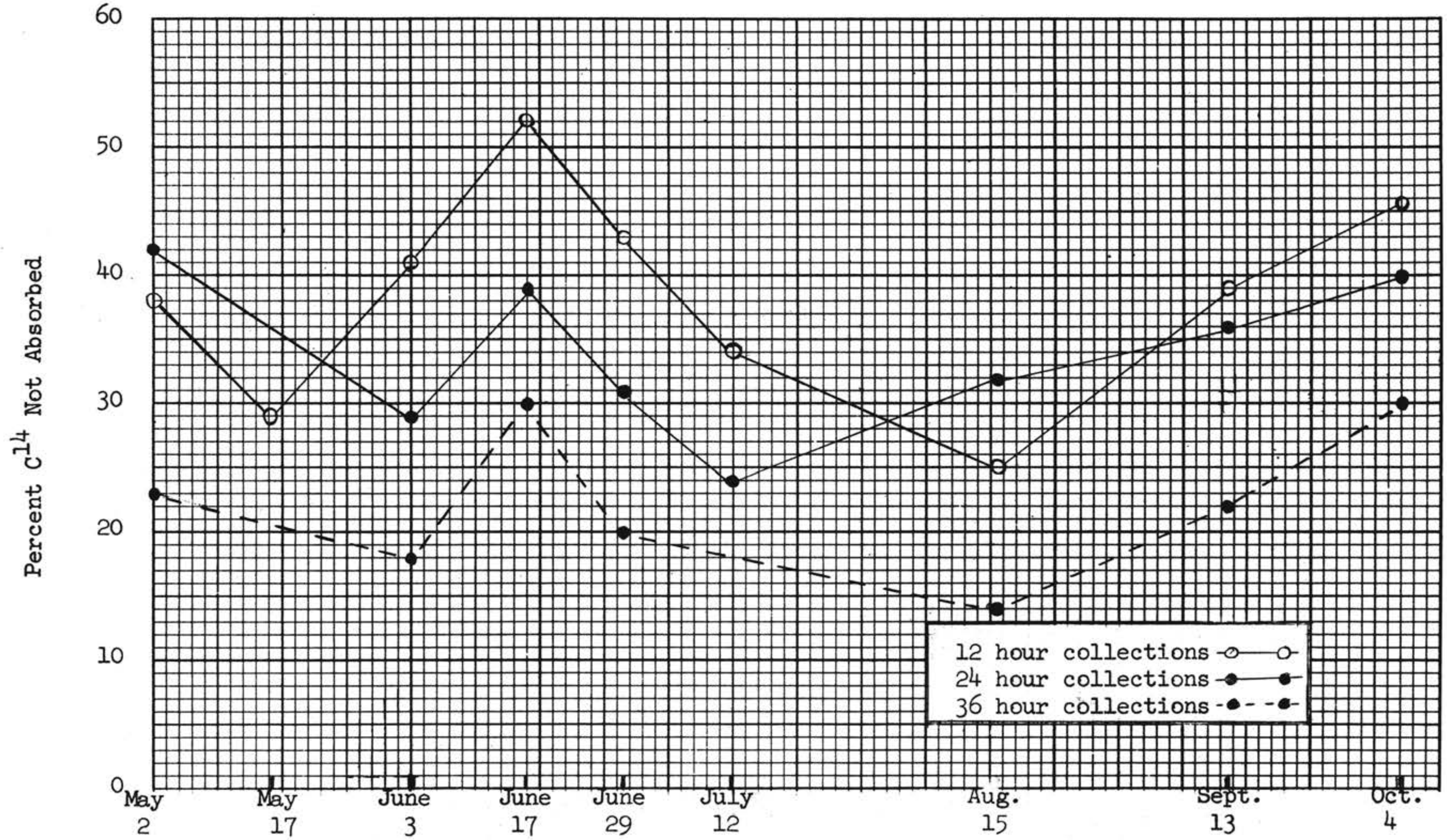
TABLE VI

DAILY RAINFALL IN INCHES AT THE OUTDOOR HYDRAULIC LABORATORY, STILLWATER, OKLAHOMA FROM JANUARY 1 TO NOVEMBER 1, 1960

	Month									
Day	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.
1	0.05	0.01				0.08				
2			0.28							
3							0.30			
4		1.32			0.34		2.35			
5	0.11									
6					1.01	1.33				
7						0.10	0.12			
8			0.04	0.10	0.13					
9								T*		
10			0.02							
11										
12	0.13					T				0.03
13				0.30			0.27			0.09
14	0.14			0.03						
15		0.25	0.24	0.09						
16	0.15			0.16						
17							1.34			
18					0.43			0.98		2.60
19										
20		0.25			0.59	0.23			0.09	
21							0.91			
22							0.07			
23		0.09							0.19	
24			0.06			0.23			0.98	
25					0.88					0.25
26								0.40	T	
27		0.20		0.36	0.20					
28				0.26	1.22					0.18
29			0.20				0.06			1.92
30										0.31
31			0.30							
Totals	0.58	2.12	1.14	1.30	5.66	2.10	5.42	2.36	0.28	5.38
Average **	1.26	1.34	1.83	2.98	4.28	4.14	2.84	3.24	3.20	2.41

* Trace

**Average monthly rainfall 2 miles west of Stillwater as computed from Oklahoma Climatological Data, Vol. 69 (13) 1960.



Appendix Figure 1. Percent of the total applied C^{14}-labeled 2,4,5-T washed from the leaf at 12, 24 and 36 hours after foliar applications to blackjack oak. The individual values represent averages for six determinations.

TABLE VII

ANALYSIS OF VARIANCE OF THE 2,4,5-T-I-C^{14} 1/ NOT ABSORBED BY BLACKJACK OAK AT 12, 24 AND 36 HOURS AFTER APPLICATION TO THE LEAF TISSUE

Source of Variation	D.F.	Sum of Squares	Mean Square	F Value
Total	125	24,227.46		
Dates	6	3,227.43		
Linear (1)		(2.74)	2.74	
Quadratic (1)		(500.30)	500.30	3.16
Cubic (1)		(1,182.92)	1,182.92	7.47*
Quartic (1)		(8.72)	8.72	0.05
Residual (2)		(1,532.74)	766.53	4.84*
Trees in Dates	14	2,217.65	158.40	
Collection Periods	2	7,510.21		
Linear (1)		(6,992.48)	6,992.48	117.67**
Quadratic (1)		(517.72)	517.72	8.71**
Collection Periods X Dates	12	1,225.11	102.09	1.71
Collection Periods X Trees in Dates	28	1,663.78	59.42	
Sample Duplicates	63	8,383.28		

1/ Determined from the percent of C^{14} washed from the leaf surface.

* Indicates significance at the 5% level of confidence.

** Indicates significance at the 1% level of confidence.

TABLE VIII

APPARENT KILL[1] OF BLACKJACK OAK WITH 2,4,5-T APPLIED AS A FOLIAR SPRAY AT VARIOUS DATES DURING THE 1960 GROWING SEASON

Treatment Date	Percent Defoliation			
	Individual Plant Evaluations[2]			Average
May 2	20	90	90	66
May 17	80	90	80	83
June 3	100	80	100	93
June 17	50	40	70	63
June 29	95	95	95	95
July 12	95	50	90	78
Aug. 15	20	0	50	23
Sept. 13	20	20	0	13
Oct. 4	60	90	30	60

1/ Apparent kill based on the percent defoliation.

2/ Made May 18, 1961.

VITA

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Master of Science

Thesis: THE EFFECT OF LEAF AGE AND ABSCISSION ZONE FORMATION ON THE ABSORPTION AND TRANSLOCATION OF 2,4,5-TRICHLOROPHENOXYACETIC ACID BY BLACKJACK OAK (QUERCUS MARILANDICA MUENCHH.)

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